Effect of Habitat Depth on Host Location by Five Species of Parasitoids (Hymenoptera: Pteromalidae, Chalcididae) of House Flies (Diptera: Muscidae) in Three Types of Substrates

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ABSTRACT Four species of pteromalid parasitoids [*Muscidifurax raptor* Girault & Sanders, *Spalangia cameroni* Perkins, *Spalangia endius* Walker, *Spalangia gemina* Boucek, and the chalcidid *Dirhinus himalayanus* (Masi)] were evaluated for their ability to locate house fly pupae at various depths in poultry manure (41% moisture), fly rearing medium (43% moisture), and sandy soil (4% moisture) from a dairy farm. Searching activity in manure was largely confined to the surface (*M. raptor*, *D. himalayanus*, and *S. gemina*) or to depths of up to 2 cm below the surface (*S. endius*, *S. cameroni*). *S. cameroni* was the most effective species at locating buried pupae in manure. All of the species searched over a wider range of habitat depths in fly rearing medium, although *M. raptor* and *S. gemina* tended to concentrate their searching activity relatively close to the surface of the substrate. Host attacks by these species at 6 cm were 30–40% lower than on the surface of the medium. *S. endius* searched uniformly at all depths in rearing medium and *S. cameroni* had highest rates of host attacks 1–2 cm below the surface of this substrate. The parasitoids displayed considerable fidelity to their search patterns regardless of whether or not they were given a choice of habitat depths in which they could find pupae. None of the parasitoids were effective at attacking fly pupae that were buried in sandy soil at any depth. The results suggest that fly larvae that pupate in the sandy soils typical of Florida's coastal plains are relatively impervious to attack by pupal parasitoids.

KEY WORDS *Musca domestica*, *Muscidifurax raptor*, *Spalangia gemina*, *Spalangia cameroni*, *Spalangia endius*, *Dihrinus himalayanus*

PUPAL PARASITOIDS IN the family Pteromalidae are important natural enemies of muscoid flies on livestock and poultry farms throughout the world (Rutz and Patterson 1990). Augmentative releases of parasitoids, especially in the genera *Muscidifurax* and *Spalangia*, can be an effective house fly management tool when used in integrated management programs (Rutz and Axtell 1979, Cabrales et al. 1985, Morgan and Patterson 1990, Geden et al. 1992, Petersen et al. 1992, Petersen and Cawthra 1995, Crespo et al. 1998). In some cases, disappointing results with parasitoid releases have been attributed to selection of appropriate species for the target fly habitat (Rutz and Axtell 1980, Petersen et al. 1983). Surveillance to determine the most important local species can be conducted to guide the selection process, but this is labor-intensive and can give different results in different years (Smith and Rutz 1991b, Jones and Weinzierl 1997). A better understanding of parasitoid niche characteristics could be helpful for matching appropriate parasitoid species to different fly breeding habitats.

Little is known about the effect of microhabitat characteristics on foraging behavior of filth fly parasitoids. Field studies have shown that parasitism rates vary in different fly breeding habitats (Petersen and Meyer 1983; Rueda and Axtell 1985a; Smith and Rutz 1991c, 1991d), and substrate moisture and light levels can affect the microhabitat choices made by different species of parasitoids (Smith and Rutz 1991a, Geden 1999). The depth at which parasitoids concentrate their search effort is an another important ecological characteristic that may vary among parasitoid species. Legner (1977) examined the depth at which eight parasitoid strains foraged in wheat flakes in small vials and concluded that *Muscidifurax uniraptor* and *M. zaraptor* concentrated their efforts near the substrate surface, whereas *Spalangia* spp. (*S. endius*, *S. nigra*, *S. cameroni*, *S. longepetiolata*) were more effective at locating buried hosts. He also noted that the search patterns of some species were affected by the moisture levels of the substrate. For example, the *Muscidifurax* spp. and *Sphegigaster* sp. were more successful in penetrating dryer than wetter media to locate buried host pupae, whereas the *Spalangia* spp. search patterns were similar at different moisture levels (Legner 1977). Although these tests provided useful comparisons of different species searching behavior, their utility for predicting behavior in the field was limited

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because of the type of substrate used (clean wheat flakes) and because the parasitoids were restricted to single burial treatments in the bioassays. King (1997) reported that *S. cameroni* was more effective than *M. raptor* at locating buried host pupae in used fly rearing medium.

Rueda and Axtell (1985b) conducted extensive field sampling in two types of poultry houses to examine the relative abundance of parasitized pupae of six parasitoid species at various depths in poultry manure. Most (>95%) of the pupae parasitized by *M. raptor*, *M. zaraptor*, and *Pachycrepoideus vindemmiae* were found within the top 3 cm of the manure surface, whereas pupae parasitized by *S. cameroni*, *S. endius*, and *S. nigroaenea* were most often collected 3–10 cm beneath the surface.

In an effort to increase the efficacy of filth fly biocontrol, we have been evaluating exotic parasitoids as candidates for possible importation and release, especially in the southern United States. These exotics include two pupal parasitoids: *Spalangia gemina* Boucek, a tropical pteromalid species collected in Brazil, and a Moroccan isolate of the chalcidid *Dirhinus himalayanus* (Masi). This evaluation has included comparisons of temperature-dependent development rates, rates of host attacks and fecundity, and the effect of habitat (poultry manure) moisture on host location (Geden 1996, 1997, 1999). The objectives of the current study were to address the following questions about host-location behavior in relation to habitat depth: (1) How do the exotic species *D. himalayanus* and *S. gemina* compare with the native species *Muscidifurax raptor*, *S. cameroni*, and *S. endius*, with respect to locating pupae at different habitat depths? (2) Does the type of substrate (poultry manure, soil, or fly rearing medium) affect parasitoid searching behavior? (3) Do parasitoids adjust their searching strategy when given a choice of depths at which hosts are present?

Materials and Methods

Parasitoid Colonies. Five species of pteromalid parasitoids were tested. *Muscidifurax raptor* Girault & Sanders and *Spalangia cameroni* Perkins were from a colony established in 1992 from a poultry farm in Brooksville, FL. The *Spalangia endius* Walker colony was established in 1994 from a poultry farm in Zephyr Hills, FL; *Spalangia gemina* Boucek was originally collected from a poultry farm near Sao Paulo, Brazil, in 1991. *Dhirinus himalayanus* (Masi) was collected from house fly pupae in Morocco in 1990. Parasitoids were maintained on ≈1- to 2-d-old house fly puparia in rearing rooms maintained at 27°C, 70–80% RH, under constant light conditions.

Substrates. Three substrates were used in the tests. Poultry manure for the bioassays was collected by placing Plexiglas panels on the surface of the manure rows in a high-rise caged layer house in Brooksville, FL. Fresh manure accumulating on the panels was collected after 24 h and frozen to kill any arthropods present. After determining that the moisture content of the fresh manure was 74%, pans of manure were partially dried in an oven at 45°C to achieve a moisture level of 41%. Moisture determinations of field-collected manure had indicated that this moisture level was similar to that of the dry manure on the manure row margins in the field where naturally occurring fly pupae were present in greatest abundance (unpublished data).

The second substrate was house fly larval rearing medium (Hogsette 1992) that had been used for rearing fly immatures to the pupal stage. The resulting "spent" medium (moisture content, 39%) was frozen within 1 h of separation of fly pupae and frozen in airtight plastic containers for up to 2 mo before testing. The third substrate was sandy loam soil (moisture content, 4%) collected from under calf bedding at a dairy farm in Bell, FL, with a history of high fly populations. The soil was sifted to remove any fly pupae present and frozen for up to 2 mo before being used in the tests.

Bioassays. Two types of bioassays were conducted to determine the ability of parasitoids to locate pupae at different substrate depths under choice and no-choice situations. In the first type of bioassay (no-choice situation), 1- to 2-d-old live fly pupae were placed in small fiberglass screen bags (50 pupae per bag) in each of the three substrates at either 0 (on the surface), 1, 2, 4, or 6 cm from the top of the substrate in 350-ml plastic cups with screen lids. The height of the substrate column was held constant at 7 cm (flush with the top of the assay cup) for all burial treatments. Five female parasitoids (2–4 d old) were introduced into each cup and the cups were covered with screened lids and held at 27°C, 70–85% RH, under constant light (3 cups per depth per substrate). Pupae were removed from the cups after 24 h, separated from any parasitoids present, and transferred to 30-cm^3 cups with snap-on screen lids for fly and parasitoid emergence. Control fly emergence was assessed by placing fly pupae in cups without parasitoids at depths of either 0 or 6 cm from the substrate surface (3 cups each) per test date. The purpose of this bioassay was to assess host location and parasitism by parasitoids when they were presented with pupae buried at a single depth in each substrate.

In the second type of bioassay (choice assays), fly pupae were first placed in the substrates at different depths as before (3 cups per depth), and the cups were placed in an array in a Plexiglas box (one type of substrate per box) measuring 55 cm long by 25 cm high by 37 cm wide. The substrate cups were arranged in three rows of 5, with each pupal depth treatment represented randomly in each row. Seventy-five female parasitoids (2–4 d old) were released in the box by placing them in an open 30-ml glass vial placed on a platform suspended from the ceiling of the box 15 cm above the cup array. In this way, the host:parasitoid ratio used in the choice bioassays tests (75 females with 750 pupae) was the same as that used in the no-choice assays (5 females with 50 pupae). Pupae were removed from the cups and transferred to 30 cm^3 cups after 24 h of exposure to the parasitoids as before.

Controls were handled as in the no-choice assays by placing pupae at either 0- or 6-cm depths in the substrates in the absence of parasitoids. These assays were conducted to evaluate the ability of the parasitoids to locate pupae presented simultaneously at a variety of substrate depths.

The entire experiment was replicated twice for each substrate, species of parasitoid, and type of bioassay (choice versus no-choice). Choice and no-choice tests for a given substrate and species were conducted on the same days to facilitate evaluation of the effect of parasitoid choice on foraging behavior. Data on numbers of attacked pupae (pupae that did not produce flies) and progeny production were analyzed by separate analysis of variance (ANOVA) for each substrate and parasitoid species using depth, choice and depth × choice as model effects using the general linear models procedure of the Statistical Analysis System (SAS Institute 1987). Data were normalized before ANOVA by a square-root transformation (Sokal and Rohlf 1981), but untransformed values were used to present results in tables. Control mortality, which averaged 3–16%, was used to calculate corrected percent host mortality (Abbott 1925) for presentation of data in tables but not used in conducting the ANOVAs. No significant differences in control mortality were observed between the 0 and 6 cm pupal burial treatments, and pooled control mortality from the two burial treatments was used in the calculation of corrected mortality.

Results

Dirhinus himalayanus attacked (killed) 30–35 of the pupae placed on the surface of poultry manure and produced 19–21 progeny per set of 50 host pupae regardless of whether other burial depth treatments were present (Table 1). Host attacks and parasitism by this species in manure was lower among pupae buried 1 cm below the surface compared with the surface pupae, but the decline was stronger when parasitoids were given a choice of burial treatments from which to select (significant depth × choice ANOVA term, Table 2). Host attacks (2–9 pupae killed) and progeny production (0.2–6.0 progeny) were low at depths of greater than 1 cm in manure. In fly larval rearing medium, host attacks (14–28) and parasitism (9–21) were more evenly distributed across depths, but there was still a tendency for somewhat higher activity at shallower pupal depths (significant only for progeny production, Table 2). In soil, nearly all of the activity was restricted to pupae placed directly on the surface regardless of the choice treatment.

In no-choice bioassays with manure, females of *M. raptor* were only effective at locating hosts that were present on the surface (47.0 pupae killed, 35 progeny produced), with negligible rates of host attacks and progeny produced at greater depths (Table 3). Females that were presented with a range of burial treatments located nearly all of the pupae on the manure surface but also killed (22.4 attacks) and parasitized (15.7 progeny) substantial numbers at 1 cm beneath the surface (significant depth × choice interaction, Table 2). Small numbers of hosts were attacked and parasitized at depths greater than 1 cm in this substrate. In fly rearing medium, *M. raptor* was most effective at locating pupae near the surface (36–48 pupae attacked in the top 1 cm) under both choice and no-choice conditions, but substantial numbers of pupae were killed in this substrate even at the maximum depth of 6 cm. Attack rates at 6 cm were about half of those observed on the surface of the medium. There was no significant change in *M. raptor*s foraging behavior in rearing medium when the parasitoids were given a range of burial treatments (nonsignificant *F* for choice, Table 2). Rates of host attack and parasitism on the surface of soil was similar to surface attack/parasitism rates in the other substrates, but essentially none of the buried pupae were attacked at (Table 3).

Spalangia cameroni was most effective at locating host pupae in the top 2 cm of poultry manure under both choice and no-choice test conditions (Table 4), and the highest rates of host attacks (40–48 pupae killed) and progeny production (26–30 progeny produced) were observed among pupae located on the manure surface. In fly larval rearing medium this species foraged more uniformly throughout the substrate column and easily located pupae buried 6 cm beneath the surface compared with manure and soil. Highest rates of host attacks were observed among pupae buried either 1 cm (37.4 pupae killed in choice assays) or 2 cm (35.2 pupae killed in no-choice assays) beneath the medium surface. Host location in sandy soil was restricted to pupae that were placed on the surface.

Spalangia endius had highest rates of host location (30–46 pupae killed, 12–26 progeny produced) in manure when pupae were either located on the manure surface or at a depth of 1 cm (Table 5), and

Table 1. Rates of host attacks and progeny production by *Dirhinus himalayanus* when house fly pupae were placed at different depths in poultry manure, fly larval rearing medium, and soil

Habitat	Depth, cm	Parasitoids not given a choice of habitat depths		Parasitoids given a choice of habitat depths	
		No. pupae killed[a]	No. parasitoids produced[b]	No. pupae killed	No. parasitoids produced
Manure	0	29.5 (3.4)	18.7 (2.8)	35.3 (3.5)	21.3 (3.7)
	1	21.3 (2.9)	9.5 (2.6)	2.9 (2.0)	0.7 (0.5)
	2	8.5 (3.6)	1.8 (1.1)	6.0 (3.0)	2.0 (2.0)
	4	6.0 (4.0)	3.0 (2.2)	2.4 (0.7)	0.2 (0.2)
	6	8.4 (3.2)	4.3 (1.9)	5.8 (5.3)	3.7 (1.7)
Medium	0	23.2 (2.0)	13.8 (1.7)	28.4 (4.1)	20.8 (4.0)
	1	22.6 (2.3)	15.7 (1.3)	25.4 (4.5)	17.5 (3.7)
	2	19.9 (4.4)	12.8 (3.0)	15.9 (4.6)	10.2 (3.2)
	4	19.4 (4.9)	11.3 (2.7)	14.0 (4.5)	9.2 (3.6)
	6	17.3 (2.2)	11.2 (1.5)	19.5 (4.5)	10.7 (2.9)
Soil	0	18.4 (2.7)	13.8 (2.7)	21.8 (2.8)	12.8 (2.4)
	1	0.9 (0.5)	0.2 (0.2)	0.9 (0.8)	0.2 (0.2)
	2	1.0 (0.3)	0.2 (0.2)	1.0 (0.5)	0.0 (0.0)
	4	0.9 (0.6)	0.0 (0.0)	1.1 (0.6)	0.0 (0.0)
	6	0.2 (0.1)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)

[a] Mean (±SE) number uneclosed pupae per assay cup of 50 pupae.
[b] Mean (±SE) number parasitoid progeny produced per assay cup of 50 pupae.

Table 2. ANOVAs for effects of habitat depth and whether parasitoids were given a choice of habitat depths on rates of host attacks and progeny production by five species of pteromalids

Species and Substrate	ANOVA *F* for host attacks			ANOVA *F* for progeny production		
	Depth[a]	Choice[b]	Depth X Choice[c]	Depth	Choice	Depth X Choice
D. himalayanus						
manure	23.25**	4.06*	3.43*	21.4**	1.64NS	1.71NS
medium	2.07NS	0.10NS	0.68NS	2.66*	0.14NS	0.91NS
soil	87.3**	0.66NS	0.71NS	55.3**	0.11NS	0.07NS
M. raptor						
manure	119.25**	12.93**	5.80**	98.04**	8.09**	4.94**
medium	9.29**	1.97NS	0.40NS	8.52**	1.51NS	1.63NS
soil	2655.44**	0.04NS	0.72NS	843.83**	1.01NS	0.73NS
S. cameroni						
manure	78.73**	1.08NS	2.72NS	80.85**	1.34NS	1.74NS
medium	3.48*	0.72NS	2.43NS	3.41*	0.25NS	2.53NS
soil	755.71**	0.02NS	0.16NS	177.2**	1.24NS	1.13NS
S. endius						
manure	70.44**	0.03NS	0.10NS	95.38**	0.15NS	1.56NS
medium	5.28**	20.70**	1.91NS	8.38**	5.86*	1.77NS
soil	86.45**	5.45*	1.74NS	107.99**	17.41**	1.52NS
S. gemina						
manure	98.82**	0.61NS	0.70NS	71.93**	2.22NS	2.07NS
medium	5.14**	0.02NS	0.34NS	4.85**	1.08NS	1.11NS
soil	95.82**	3.75NS	2.11NS	113.42**	4.11*	2.93**

[a] Pupae buried 0, 1, 2, 4, or 6 cm beneath substrate surface (d = 4, 50).
[b] Choice versus no-choice bioassays (df = 1, 50).
[c] d = 1, 50.

responses were similar under both choice and no-choice test conditions (Table 2). In fly larval rearing medium this species foraged throughout the substrate but had significantly (Table 2) higher attack and parasitism rates at greater depths than near the surface of this substrate. Activity in soil was restricted to pupae that were placed on the surface.

Spalangia gemina's search pattern in manure resembled that of *S. cameroni*, with most activity observed in the top 2 cm of the manure under both choice and no-choice test conditions (Table 6). In rearing medium this species foraged throughout the substrate but resembled *M. raptor* and *D. himalayanus* by being more active closer to the surface than at greater depths in this substrate. Activity in soil was restricted to pupae that were placed on the surface.

Table 3. Rates of host attacks and progeny production by *Muscidifurax raptor* when house fly pupae were placed at different depths in poultry manure, fly larval rearing medium, and soil

Habitat	Depth, cm	Parasitoids not given a choice of habitat depths		Parasitoids given a choice of habitat depths	
		No. pupae killed[a]	No. parasitoids produced[b]	No. pupae killed	No. parasitoids produced
Manure	0	47.0 (2.2)	35.0 (1.3)	49.1 (0.6)	35.0 (1.4)
	1	1.1 (0.5)	0.0 (0.0)	22.4 (6.5)	15.7 (5.7)
	2	0.5 (0.4)	0.0 (0.0)	5.0 (4.3)	2.7 (2.7)
	4	0.8 (0.6)	0.0 (0.0)	0.7 (0.7)	0.2 (0.2)
	6	1.8 (0.8)	0.0 (0.0)	3.3 (1.4)	0.7 (0.7)
Medium	0	48.1 (1.2)	37.7 (1.6)	47.1 (2.6)	41.3 (2.5)
	1	44.2 (1.9)	36.2 (3.1)	36.1 (5.2)	27.0 (5.8)
	2	38.3 (3.6)	31.2 (2.8)	35.8 (3.2)	26.8 (3.0)
	4	32.5 (4.9)	24.2 (4.5)	32.8 (4.8)	29.0 (5.1)
	6	27.2 (5.6)	22.5 (5.1)	20.1 (5.4)	12.5 (2.9)
Soil	0	48.8 (0.6)	39.7 (2.2)	49.2 (0.4)	37.3 (1.5)
	1	0.3 (0.3)	0.2 (0.2)	0.9 (0.7)	0.0 (0.0)
	2	0.0 (0.0)	0.0 (0.0)	0.3 (0.2)	0.0 (0.0)
	4	0.2 (0.2)	0.2 (0.2)	0.7 (0.7)	0.0 (0.0)
	6	1.5 (1.3)	0.0 (0.0)	0.3 (0.2)	0.0 (0.0)

[a] Mean (±SE) number uneclosed pupae per assay cup of 50 pupae.
[b] Mean (±SE) number parasitoid progeny produced per assay cup of 50 pupae.

Discussion

Previous studies have indicated that *Muscidifurax* spp. tend to be superficial searchers that locate pupae near the surface of test substrates, whereas *Spalangia* spp. have a tendency to search at greater depths (Legner 1977, Rueda and Axtell 1985b, King 1997). Results presented here support those generalizations to a degree, but the physical properties of various substrates clearly impose limits on the parasitoids' innate preferences. In the most challenging substrate, sandy loam soil, none of the parasitoids were effective at locating pupae that were buried even by a very thin (1 cm) layer of the substrate. In poultry manure we found that all of the species tested were primarily superficial or subsurface foragers. Although the manure in these tests had been dried to 43% moisture, it may have had other physical characteristics (e.g., particle size) that inhibited parasitoids from penetrating more than a few cm below the surface. Under field conditions, species that prefer to forage at greater depths may be limited to surface regions in dense substrates such as soil, compacted silage or wet manure. It is also possible that naturally occurring pupae below the surface are easier for parasitoids to locate than our experimentally placed pupae because of semiochemical trails and break points in the substrates that may be left by the larvae before pupation.

Table 4. Rates of host attacks and progeny production by *Spalangia cameroni* when house fly pupae were placed at different depths in poultry manure, fly larval rearing medium, and soil

Habitat	Depth, cm	Parasitoids not given a choice of habitat depths		Parasitoids given a choice of habitat depths	
		No. pupae killed[a]	No. parasitoids produced[b]	No. pupae killed	No. parasitoids produced
Manure	0	40.1 (2.3)	26.0 (1.0)	48.4 (0.3)	30.3 (1.4)
	1	18.2 (2.3)	11.2 (1.1)	27.3 (6.6)	16.0 (3.8)
	2	15.7 (3.7)	7.8 (2.5)	9.2 (3.3)	4.3 (2.6)
	4	1.9 (1.1)	0.0 (0.0)	1.4 (0.9)	0.8 (0.8)
	6	1.4 (0.3)	0.7 (0.7)	0.4 (0.2)	0.8 (0.8)
Medium	0	20.8 (1.1)	12.7 (1.1)	29.0 (4.1)	17.0 (1.6)
	1	33.2 (3.1)	18.8 (2.3)	37.4 (1.8)	23.3 (1.4)
	2	35.2 (2.6)	21.8 (1.9)	27.0 (5.4)	15.2 (3.1)
	4	30.5 (4.0)	16.3 (2.7)	17.6 (5.1)	10.0 (2.4)
	6	24.5 (2.8)	15.0 (2.2)	22.7 (6.1)	15.3 (4.2)
Soil	0	45.1 (2.0)	30.2 (2.5)	45.7 (2.5)	25.7 (3.4)
	1	0.6 (0.5)	0.2 (0.2)	0.1 (0.1)	0.0 (0.0)
	2	0.0 (0.0)	0.0 (0.0)	0.4 (0.3)	0.0 (0.0)
	4	0.3 (0.2)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)
	6	0.8 (0.4)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)

[a] Mean (±SE) number uneclosed pupae per assay cup of 50 pupae.
[b] Mean (±SE) number parasitoid progeny produced per assay cup of 50 pupae.

Table 6. Rates of host attacks and progeny production by *Spalangia gemina* when house fly pupae were placed at different depths in poultry manure, fly larval rearing medium, and soil

Habitat	Depth, cm	Parasitoids not given a choice of habitat depths		Parasitoids given a choice of habitat depths	
		No. pupae killed[a]	No. parasitoids produced[b]	No. pupae killed	No. parasitoids produced
Manure	0	45.0 (1.3)	28.7 (2.3)	47.7 (1.0)	25.0 (3.0)
	1	11.8 (4.3)	3.7 (1.2)	17.3 (6.0)	6.5 (2.7)
	2	7.3 (2.7)	1.7 (0.8)	4.2 (2.2)	0.0 (0.0)
	4	0.9 (0.4)	0.2 (0.2)	1.2 (0.6)	0.0 (0.0)
	6	1.2 (0.4)	0.0 (0.0)	2.4 (1.1)	0.0 (0.0)
Medium	0	46.6 (1.5)	33.5 (3.7)	46.7 (1.2)	39.2 (2.6)
	1	36.3 (4.4)	26.3 (3.6)	39.4 (4.0)	31.5 (3.5)
	2	31.6 (5.8)	24.2 (5.0)	24.2 (5.4)	15.2 (4.3)
	4	29.3 (5.8)	17.3 (4.9)	29.2 (5.6)	18.5 (5.0)
	6	29.0 (6.5)	18.8 (5.0)	30.7 (6.1)	24.6 (6.4)
Soil	0	23.1 (4.2)	19.5 (3.6)	31.2 (2.5)	27.2 (2.2)
	1	0.3 (0.3)	0.0 (0.0)	1.9 (1.9)	1.2 (1.2)
	2	0.3 (0.2)	0.0 (0.0)	2.0 (0.9)	0.0 (0.0)
	4	1.0 (0.5)	0.0 (0.0)	0.7 (0.3)	0.0 (0.0)
	6	0.8 (0.4)	0.0 (0.0)	0.1 (0.1)	0.0 (0.0)

[a] Mean (±SE) number uneclosed pupae per assay cup of 50 pupae.
[b] Mean (±SE) number parasitoid progeny produced per assay cup of 50 pupae.

The results with rearing medium were similar to those of Rueda and Axtell (1985b) for the three species that the two studies had in common; *M. raptor*, *S. endius*, and *S. cameroni*. *M. raptor* foraged to depths of 5–6 cm with little difficulty in both studies, although proportionally greater activity was observed closer to the surface. Similarly, *S. endius* and *S. cameroni* in both studies displayed a tendency to search across a wide range of depths. We observed considerably higher rates of attacks on the surface by *Spalangia* spp. than were observed by Rueda and Axtell (1985). The latter authors hypothesized that the *Spalangia* spp. may seek pupae at greater depths to avoid superparasitism encounters with *Muscidifurax* spp., who are generally the victors in such encounters (Propp and Morgan 1983). If this is the case, then *Spalangia* spp. may search surface regions more thoroughly in the absence of heterospecific competitors. Indeed, King (1997) found that *S. cameroni* reproduction on buried hosts was higher when *M. raptor* were included in the assay arenas. It is possible that *Spalangia* spp. under field conditions can detect the presence of competitively superior *Muscidifurax* spp. and refine their search patterns to minimize competition events that would work to their disadvantage. Further research would be required to evaluate the impact of the presence of heterospecific competitors on the search patterns of these parasitoids.

Table 5. Rates of host attacks and progeny production by *Spalangia endius* when house fly pupae were placed at different depths in poultry manure, fly larval rearing medium, and soil

Habitat	Depth, cm	Parasitoids not given a choice of habitat depths		Parasitoids given a choice of habitat depths	
		No. pupae killed[a]	No. parasitoids produced[b]	No. pupae killed	No. parasitoids produced
Manure	0	43.6 (2.7)	28.5 (3.5)	45.5 (1.8)	23.3 (1.4)
	1	33.1 (5.9)	10.0 (2.5)	32.5 (4.7)	12.8 (2.6)
	2	6.4 (2.3)	0.0 (0.0)	5.4 (1.1)	0.3 (0.2)
	4	9.4 (3.1)	0.0 (0.0)	7.6 (2.4)	0.0 (0.0)
	6	4.2 (2.1)	0.0 (0.0)	4.0 (1.6)	0.0 (0.0)
Medium	0	45.5 (0.9)	26.3 (2.0)	33.0 (4.7)	12.3 (3.4)
	1	44.5 (2.5)	24.2 (4.1)	30.4 (4.9)	21.2 (4.5)
	2	48.5 (0.7)	32.8 (2.7)	45.8 (2.0)	31.5 (2.3)
	4	47.8 (0.7)	31.8 (1.2)	45.2 (3.0)	31.2 (2.1)
	6	48.9 (0.7)	32.7 (2.5)	41.5 (2.7)	29.5 (2.5)
Soil	0	27.0 (1.8)	16.0 (2.0)	17.8 (2.2)	6.8 (0.9)
	1	0.9 (0.8)	0.0 (0.0)	1.6 (0.7)	0.0 (0.0)
	2	1.3 (0.7)	0.0 (0.0)	1.2 (0.8)	0.0 (0.0)
	4	1.2 (0.8)	0.0 (0.0)	1.1 (0.6)	0.0 (0.0)
	6	1.3 (0.6)	0.0 (0.0)	1.7 (0.7)	0.0 (0.0)

[a] Mean (±SE) number uneclosed pupae per assay cup of 50 pupae.
[b] Mean (±SE) number parasitoid progeny produced per assay cup of 50 pupae.

Spalangia gemina displayed search pattern characteristics that were most similar to those of *S. cameroni*. These two species resemble each other morphologically and in their manure moisture preferences, development times, and attack rates (Geden 1996 1997, 1999). Little is known about the biology of this tropical species in the field, but it has been found parasitizing house flies on poultry farms in Brazil sympatrically with *S. cameroni* at the same times of year (Ferreira de Almeida and Pires do Prado 1998). The other exotic species tested, *D. himalayanus*, attacks a wide range of hosts and has lower overall attack rates than the other species studied here (Geetha-Bai 1990). This species appears to be similar to *M. raptor* with regard to searching behavior as a function of habitat depth (this study) and manure moisture preferences (Geden 1997). No information is available on the outcome of competition between these two species, but it is likely

that *M. raptor* would overwhelm *D. himalayanus* in the field because of its faster development rate and aggressive larvae.

None of the parasitoids were able to forage below the surface of sandy loam soil from a dairy farm. Large numbers of pupae were present in the soil when it was collected and had to be removed before the soil was used in the tests. In regions with extensive coastal plains such as Florida, fly pupae can be found 15–30 cm below the soil surface near fly breeding areas (Hogsette 1996). Our data suggest that such pupae are well protected from parasitoid attacks, although field surveys for pupal parasitism at different depths in soil are needed to confirm this.

Most of the species tested, especially the *Spalangia* spp., displayed considerable fidelity in their search patterns and located pupae in different depth zones at similar rates regardless of whether they were in choice or no-choice situations. This was surprising because it was anticipated that the parasitoids would be more willing to search outside their preferred zones if the only hosts available to them were outside those zones (no-choice tests). The only time that this hypothesis was borne out was in the manure tests with *D. himalayanus*, which located more pupae below the surface in no-choice tests than it did in tests where the parasitoids were presented with a range of host burial treatments. In a related study, the same five species of parasitoids were found to be comparatively flexible in their manure moisture preferences; females would switch over to less-preferred moisture zones when hosts in the preferred zones were in short supply (Geden 1999).

In summary, the five species tested showed distinct search patterns with regard to habitat depth, but these patterns were modulated by the type of substrate tested. Because fly pupae can occur in a wide range of habitats within individual farms, the results also support the suggestion of Rueda and Axtell (1985b) that releases of multiple species with complimentary foraging behaviors such as *M. raptor* and *S. cameroni* might be more effective than single-species releases for fly management programs.

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